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(54) **Modified cereal flour and processed food using the same**

(57) This invention provides a modified cereal flour such as wheat flour imparted with excellent quality through modification, and additionally provides a cereal-flour processed food with excellent quality such as elasticity, smoothness and extra-fineness, produced from the modified cereal flour as a raw material. This is achieved by a method for modifying cereal flour by transglutaminase treatment during the process of milling cereal flour. The invention also provides cereal-flour based processed foods such as noodles, breads, pastries and the like produced from the modified cereal flour as the raw material.

Description**Industrial Field of the Invention**

5 The present invention relates to a modified cereal flour and a cereal-flour based processed food produced by using the same modified cereal flour. More specifically, the present invention relates to a modified cereal flour produced by effecting transglutaminase treatment during the process for producing cereal flour such as wheat flour from raw material cereal grains such as wheat, and various cereal-flour based processed foods including breads, pastas such as macaroni and spaghetti, Chinese noodles (including the wrappings of ravioli, wang-tang, and the like), Japanese noodles
 10 such as udon and soba, tempura, oil-fried quick breads such as doughnut, cakes, snacks, fresh pastries, and Japanese-style pastries.

Prior Art

15 As to the modification of wheat flour for use in breads, pastries and cakes, a great number of research works have been carried out, conventionally. For example,

1. a method comprising keeping wheat flour in an atmosphere of carbon dioxide gas and ethanol at 40 °C or more (Japanese Patent Publication (Kokoku) No. 6 - 36725);
- 20 2. a method comprising adding 40 to 500 % by weight of water to the raw material wheat, drying the resulting wheat at a temperature where no denaturation occurs, to prepare the wheat as cereal flour for pastries (Japanese Patent Publication (kokoku) No. 5 - 4055); and
3. a method comprising adding an oxidant and water into wheat flour, to recover gluten with excellent processability for processed food (Japanese Patent Publication (kokoku)No. 6 - 34682) and the like.

25 Furthermore, reports have been published about a technique to modify wheat flour by using transglutaminase (sometimes abbreviated as "TG" hereinafter) which is an enzyme catalyzing the acyl transfer reaction of the γ -carboxyamide group in the glutamine residue in peptides. For example, a method comprising adding a given amount of TG into commercially available wheat flour for cake to prepare wheat flour with excellent taste and texture for cake (Japanese
 30 Patent Laid-open (kokai) No. 2 - 286031) and a method comprising adding TG into commercially available wheat flour to prepare dough with good elasticity for bread preparation (USP No. 5,279,839) have been reported.

Each of the above described techniques is individually excellent in some respect. However, as described below, a technique which can essentially overcome the problems specific to wheat flour has never been reported.

More specifically, the problems are such that

- 35 1. deterioration such as oxidation of wheat flour occurs after storage for a long term;
2. wheat flour causes allergy;
3. preferable taste and texture cannot be imparted to the final products (cereal-flour processed food) such as bread, pastries, cake, pasta and the like.

Problems that the Invention is to Solve

40 Thus, it is an object of the present invention to provide a novel cereal flour, for example, wheat flour, with (1) no deterioration such as oxidation after storage for a long term and with (2) low allergenic action and (3) capable of imparting preferable taste and texture to final products, including bread, pastries, cake, pasta, etc.

Means for Solving the Problems

50 The present inventors have made investigations to overcome the problems described above. Consequently, the inventors have found that the problems can be overcome by a transglutaminase treatment in which the action of transglutaminase is exerted during the process of producing cereal flour such as wheat flour, which can be used as a material of the cereal-flour processed product. Thus, the present invention has been achieved on the basis of such finding.

More specifically, the present invention relates to a modified cereal flour obtained by effecting transglutaminase treatment at (1) the tempering process and/or (2) the grinding process for producing cereal flour from the raw material
 55 cereal grain, and a cereal-flour based processed food using such a modified cereal flour as the raw material.

As has been described above in the preceding paragraph, examples wherein transglutaminase is used to modify wheat flour commercially available, have been already known. However, in these examples, transglutaminase is used for treating the raw materials, such as wheat flour milled from wheat, for wheat-flour based processed food. These

examples are completely different from the present invention, because transglutaminase exerts its action during a process of milling as disclosed in the present invention. Needless to say, the use of transglutaminase in the course of milling is surprisingly more effective.

5 In accordance with the present invention, the aforementioned various problems can be overcome, for example, by making use of the action of transglutaminase during the process of producing wheat flour. Additionally, the gel forming capacity, viscosity and water retention capacity of wheat protein contained in wheat flour can be improved, to readily provide a modified wheat flour highly ranked as a raw material of breads, tempura, cakes, fried quick breads such as doughnut and batter powder.

10 Embodiments of Carrying out the Invention

The present invention will now be described in detail below.

The term "cereal flour" in accordance with the present invention means any one cereal flour of wheat, barley, corn, Japanese soba-ko, rye, oats, Chinese millet, and soy bean or a mixture cereal flour of two or more of these cereal flours
15 in combination. These cereal flours are generally used for producing breads, pastas such as macaroni and spaghetti, Chinese noodles (including the wrappings of gyoza, wang-tang, and the like), Japanese noodles such as udon and soba, tempura, oil-fried quick breads such as doughnut, cakes, snacks, fresh pastries, and Japanese-style pastries.

The transglutaminase to be used for producing the modified cereal flour of the present invention is an enzyme catalyzing the acyl-transfer reaction of the γ -carboxamide group in the glutamine residue in peptides. When the transglutaminase acts on the ϵ -amino group of the lysine residue in a protein as an acyl receptor, ϵ -(γ -Glu)Lys bonds are formed intramolecularly or intermolecularly, whereby the cross-linking reaction of the protein in wheat flour as a main raw material for preparing the processed food is promoted, so that a wheat flour with properties such as higher gel formation capacity, a higher viscosity and higher water retention capacity can be obtained.

For example, the detailed properties of the enzyme(transglutaminase) derived from microorganisms are described
25 in Japanese Patent Laid-open(kokai) No. 64 - 27471.

Transglutaminase is classified into calcium-non-dependent type and calcium-dependent type, and any of these types is satisfactorily used for the production of the modified cereal flour. Examples of the former include those from microorganisms (see, for example, the Japanese Patent Laid-open(Kokai) No. 64 - 27471 described above). Examples of the latter include those from liver of guinea pig (see Japanese Patent Publication(Kokoku) No. 1 - 50382), those from animal blood (also referred to as "Factor XIII"), those from fish (see, for example, Journal of Japan Fisheries Academy, Vol. 56, page 125 - 132 (1990)) and Proceedings of Nippon Fishery Federation, 1994, page 219). Besides, those produced by the method of genetic engineering can be included (see Japanese Patent Laid-open(Kokai) Nos. 1 - 300889 and 5 - 199883), and any of such transglutaminase can be used, satisfactorily, with no specific limitation to the origin and the preparation process.

For example, in the case of production of wheat flour, the amount of transglutaminase to be added (to be used) in the process of preparing the modified cereal flour is 0.01 to 100 units (U), preferably 0.1 to 50 units (U) per 1 g of the protein in the raw material wheat. If the amount thereof to be added is less than the range described above, the gel formation capacity is reduced. Also, in case where the resulting modified cereal flour is used for the production of bread, the resulting bread becomes too soft, and is unpreferable; and if the amount is above the range, alternatively, the resulting bread becomes too hard, so that the preferable taste and touch of the bread are deteriorated. Even if the types of the raw material wheat are different, the range of the amount of transglutaminase to be used is the same.

For the purpose of the present invention commercially available enzyme formulations comprising transglutaminase as a main component may be used (for example, "Activa" (specific activity of 1 U/ml) manufactured by Ajinomoto Co., Inc.).

45 In accordance with the present invention, the activity unit of the transglutaminase is assayed and defined as follows. More specifically, after the reaction of transglutaminase with benzoyloxycarbonyl-L-glutaminyglycine and hydroxylamine as substrates to form an iron complex with the resulting hydroxamic acid in the presence of chloroacetic acid, the absorbance at 525 nm is measured. In such manner, a standard curve is prepared on the basis of the amount of hydroxamic acid, and the enzyme amount that generates 1 μ mol hydroxamate per one minute is defined as 1 unit (U)
50 of the transglutaminase activity. As to the details of the assay, see the Japanese Patent Laid-open(Kokai) No. 64 - 27471.

When the transglutaminase treatment is carried out in the presence of a foreign protein and/or a partial protein hydrolysate, the effect of the present invention can be improved, such that the resulting wheat flour in a kneaded mixture with water has further increased elasticity and viscosity.

55 The term "foreign protein" means protein intentionally added extraneously for the transglutaminase treatment, excluding the protein naturally contained in the cereal grains (cereals) to be milled. Such protein includes milk protein such as casein, sodium caseinate, calcium caseinate, whole fat dry milk, and skimmed dry milk; wheat protein; gelatin; collagen; corn protein such as zein; rice protein; soy bean protein such as separated soy bean protein, extracted soy

bean protein and soy bean whey protein; egg protein such as egg white albumin; milk whey protein and the like.

Furthermore, the partial protein hydrolysate includes partially hydrolyzed products of protein to be used as the foreign proteins, such as the partially hydrolyzed products of wheat protein, milk protein and so on.

The partial protein hydrolysates obtainable by partially hydrolyzing the proteins, such as wheat protein and milk protein, by enzymes, acids and alkalis according to routine methods, can be used.

No specific limitation is imposed as long as the partial protein hydrolysate can achieve the objects of the present invention.

As the partial protein hydrolysate, furthermore, commercially available peptides such as lysine peptide and glutamine peptide may be used as the partial protein hydrolysate having the same effects, other than the partial protein hydrolysate derived from natural protein, as described above. Therefore, the partial protein hydrolysate in accordance with the present invention includes peptides such as lysine peptide composed of the single amino acid.

The average molecular weight of the partial protein hydrolysate to be used in accordance with the present invention is generally about 600 to 40,000, preferably about 3,000 to 20,000 and more preferably about 6,000 to 16,000.

The amount of the foreign protein and/or the partial protein hydrolysate to be added (to be used) is 0.001 to 2.0 g, preferably 0.01 to 1.0 g per 1 g of the protein of wheat, when the raw material cereal grain is wheat. The amount thereof to be added does not vary, whether the foreign protein or the partial protein hydrolysate is used singly or is used in combination of both the foreign protein and the partial protein hydrolysate (namely, the total amount of the two is fixed in the above-mentioned range.). If the amount thereof to be added is less than 0.001 g, the noodles produced by using the resulting wheat flour cannot get improved limpness and elasticity; or the wrappings of gyoza (ravioli), wang-tang, and the like cannot be also improved. When the amount is above 2.0 g, alternatively, noodles and the wrappings of gyoza (ravioli), wang-tang, and the like obtained by using the resulting wheat flour are disadvantageously fragile, with non-viscous texture. In either case, the initial objects cannot sufficiently be attained.

A method for producing the modified cereal flour of the present invention from the cereal grain, by using concurrently the transglutaminase described above and, if desired, a foreign protein and/or a partial protein hydrolysate, is now described in detail below.

Firstly, the concept of milling should be described for the sake of deep understanding.

As well known, milling is described as follows. "Cereals with the outer side composed of sugar layers being soft and with the inner endosperm being hard, like rice grain, are shaved from the outside, to separate the edible part (rice pearling). On the other hand, cereal flour with the outer sheath being very strong but with the endosperm being fragile and being readily broken and, additionally, with longitudinal grooves at the grain center, should be pulverized to then separate the sheath part. Such method, namely milling, is carried-out. So as to effectively utilize gluten specific to wheat, wheat is necessarily prepared as wheat flour. The milling process is summarized as follows.

1. Selection

A process of removing micro-fine contaminated matters such as stone. Because it is difficult to remove impurities from the resulting product of wheat flour, selection of raw material wheat should be done very carefully.

2. Tempering and blending

For the purpose of strengthening the sheath, separating the endosperm part easily, and softening the endosperm part for ready milling, water is added to wheat, which is then rested as it is 24 to 48 hours, for tempering. Additionally, wheat raw materials separately tempered are blended together, if necessary, depending on the object.

3. Grinding

The sheath of the tempered wheat is separated as much as possible by means of a brake roll, to recover crude grain of the endosperm (disruption process). Then, sifting the crude grain and transferring the grain to a purifier, to remove the contaminated debris of the sheath by a combination of sifting and draft selection (purification process). Furthermore, the purified crude grain is ground by means of a smooth roll (smooth face or crude face), followed by separating the flour by grain size through sifting (pulverization process).

4. Blending of wheat flour

After sifting, wheat flours of various sizes (finished flour; agari-ko in Japanese) are blended together, depending on the properties thereof, so as to prepare a wheat flour of the objective quality and grade.

5. Finishing

A final product is produced after thorough mixing. So as to supplement the product with vitamins and the like, then, vitamins and the like should be mixed into the product." (see "General Dictionary of Food Industry", new edition, issued by Korin K.K, 1993). It is needless to say that such milling is applicable to other cereal grains (cereals) with the same structure as that of wheat.

The transglutaminase treatment during the milling process will now be described below by referring to the example of wheat. Except for the treatment, the modified wheat flour of the present invention can be produced according to the well-known milling method from raw material wheat.

The preferable protein content of wheat flour varies depending on the use thereof. Additionally, the protein content of wheat flour varies depending on the type of the raw material wheat. Wheat flour is divided into gluten-rich wheat flour (grade I; kyoriki-ko in Japanese), semi-gluten-rich wheat flour (grade II; jun-kyoriki-ko in Japanese), moderately-gluten-rich wheat flour (grade III; churiki-ko in Japanese), gluten-poor wheat flour (grade IV; hakuriki-ko), durum semolina flour, and the like. In accordance with the present invention, any of the wheat flours can be modified effectively.

The transglutaminase treatment in the course of the tempering process can be performed for example as follows.

When standard Canada Western Red Spring wheat is selected as semi-gluten-rich wheat flour (grade II), transaminase is added at 0.01 to 100 U, preferably 0.1 to 50 U per 1 g of the protein of the raw material wheat during the course of water addition at the tempering process, together with a foreign protein and/or a partial protein hydrolysate at 0.001 to 2.0 g, preferably 0.01 to 1.0 g in total per 1 g of the protein of the wheat, if desired. The amount of water to be added is with no specific limitation, but generally, water is added such that the final water content in the wheat is about 10 to 20 %, preferably about 12 to 16 %. Subsequently, tempering is effected generally at 0 to 60 °C, preferably at 10 to 30°C, for 16 to 50 hours, so that transglutaminase is permeated from the surface of the wheat grain through the part of the germ into the inside thereof, to promote the crosslinking of gluten in the wheat protein. Through such tempering, the endosperm is readily pulverized, while the epidermis absorbs water moderately and furthermore gets hard due to the action of transglutaminase, so that the resulting grain becomes so fragile as to be very readily disrupted. Additionally, the gluten in the protein inside the endosperm is crosslinked, so that a wheat flour with properties such as elasticity and the capability of imparting suppleness to the resulting noodles can be recovered.

The wheat flour obtained by such transglutaminase treatment is the modified wheat flour with improved performance.

The transglutaminase treatment at the grinding process is carried out for example as follows.

Water is added to the raw material wheat after tempering and blending 1 to 3 hours prior to the grinding process.

Transglutaminase and, if desired, a foreign protein and/or a partial protein hydrolysate, are dissolved or dispersed in the water to be added during the course of water addition. The wheat to which are added transglutaminase and, if desired, the foreign protein and/or the partial protein hydrolysate is subjected to a procedure to separate the endosperm from the epidermis (disruption process), and thereafter, procedures according to routine milling process are effected to recover the modified wheat flour of the present invention. The amount of transglutaminase to be then added is 0.01 to 100 U, preferably 0.1 to 50 U, per 1 g of the protein of the raw material wheat; and the amount of the foreign protein and/or the partial protein hydrolysate to be added in total is 0.001 to 2.0 g, preferably 0.01 to 1.0 per 1 g of the protein of the raw material wheat.

The transglutaminase treatment is carried out at either of the tempering process and the grinding process as described above, but in some case, the treatment may be carried out at both the tempering process and the grinding process.

Although, furthermore the transglutaminase treatment may be effected at a process step subsequent to the grinding process, the effect is poor. For example, the transglutaminase treatment of the wheat flour (finished flour) at the blending or finish process after the grinding process is effected for example as follows.

Transglutaminase in an amount of 0.01 to 100 U, preferably 0.1 to 50 U per 1 g of the protein in the wheat flour is preliminarily dissolved in an appropriate amount of water. If desired, further, a foreign protein and/or the partial protein hydrolysate after the grinding process, at 0.001 to 2.0 g, preferably 0.01 to 1.0 g per 1 g of the wheat protein is added, together with transglutaminase, followed by dissolution or dispersion. Subsequently, the resulting solution is sprayed over the wheat flour through the pulverization process (last process step of the grinding process), and the flour is aged at 5 to 35 °C for 15 minutes to 48 hours, preferably 1 to 24 hours. During the "aging" process, the transglutaminase exerts its action. The wheat flour through the "aging" process is dried in air and finished as a product.

Such method for modifying wheat flour in accordance with the present invention is significantly different from those known methods using transglutaminase as described by the Japanese Patent Laid-open (Kokai)No. 2 - 286031 and USP No. 5,279,839, in that transglutaminase exerts its action at the milling process to produce wheat flour from wheat as an agricultural product.

A cereal-flour based processed food using the modified cereal flour of the present invention is now described below.

The method for producing the cereal-flour based processed food using the modified cereal flour of the present invention can follow conventional methods for producing processed food from cereal flour, except for the use of the modified cereal flour of the present invention as the raw material.

Among breads, for example, a loaf of bread is generated for example as follows. Yeast, yeast food and water are kneaded into the raw material modified wheat flour of the present invention by means of a mixer. Subsequently, the kneaded mixture is kept at 20 to 40 °C for 20 minutes to 10 hours for a first fermentation, to prepare intermediate seed dough. Water, edible salt, sugars, oil, skimmed dry milk and the like are added to and kneaded with the intermediate seed dough, to prepare bread dough. The bread dough is divided appropriately into portions, which are left to stand at 20 to 40 °C for a given time for the purpose of forming the network structure of wheat gluten (fermentation) and are then filled in a baking pan. Subsequently, fermentation is again progressed. The total fermentation time is about 20 minutes to 12 hours. After completion of fermentation, the dough is baked in an oven at 180 to 250 °C.

The loaf of bread obtained by baking is excellent in that the bread can maintain good taste and properties (for example, the capacity of maintaining the final shape of bread) even after long-term storage.

It is needless to say that a direct kneading process comprising kneading together all of the modified wheat flour, yeast and other raw materials prior to the aforementioned first fermentation is satisfactory to prepare a loaf of bread, besides the intermediate seed dough preparation method. The first fermentation is carried out by keeping the kneaded product at a temperature of 20 to 45 °C for 30 minutes to 10 hours. A subsequent fermentation process is done under general fermentation conditions for preparing bread. The fermentation conditions are with no specific limitation. If necessary, additionally, the bread dough may be again fermented, after resting at a temperature of 20 to 45 °C.

Instead of baking, furthermore, steamed bread can be recovered by heating in steam. It is needless to say that the heating conditions for heating in steam are according to conventional methods.

For preparing a loaf of bread, other ingredients routinely used, which include skimmed dry milk, egg, polysaccharide, fruit, coffee extract components, spice, seasoning, additives such as ascorbic acid, swelling agents (ammonium hydrogen carbonate, sodium hydrogen carbonate etc.), bleaches (ammonium persulfuric acid, potassium bromate, etc.), quality modifiers (calcium stearoyl lactate, L-cysteine hydrochloride, etc.), emulsifiers (glycerin fatty acid esters, sucrose fatty acid esters, etc.), may be used in addition to the raw material modified cereal wheat flour, yeast, yeast food, water, edible salt, sugar and fat and so on.

It is needless to say that fried quick breads such as doughnuts are prepared according to routine methods by using the modified cereal flour of the present invention as the principal component. Secondary raw materials are with no specific limitation, and therefore, sugar, egg, egg white, cream, butter, milk, seasoning, edible salt, spice and the like are appropriately used, if necessary. The fried quick breads such as doughnuts, using the modified wheat flour of the present invention, are very tasty.

It is needless to say that the preparation of cakes such as sponge cake by using the modified cereal flour of the present invention follows routine methods. Secondary raw materials are with no specific limitation, and therefore, egg, sugar, milk, butter and the like, being usually used, are appropriately used. It is needless to say that the preparation of sponge cake can be carried out by using general equipment.

For example, foamed meringue is prepared by foaming the resulting mixture which is obtained by adding sugar into egg yolk by using a Hobart mixer at a low speed for well agitation, adding simultaneously egg white and sugar into the resulting mixture.

Subsequently, the modified wheat flour of the present invention is added to and mixed with the meringue, followed by addition of melted butter if necessary and further gradual agitation at a low speed for mixing them together. The dough is poured into a pan, followed by baking on medium flame at 160 to 180 °C for about 30 minutes, thereby preparing sponge cake. The sponge cake obtained by using the modified wheat flour of the present invention presents preferable taste with elasticity.

It is needless to say that the preparation of Japanese-style pastries using the modified cereal flour of the present invention may follow general methods. In the case where the Japanese-style pastry is a type of bean pastry covered with a cherry blossom leaf (sakura-mochi), water is added, together with sugar and rice flour as secondary raw materials, into the modified wheat flour, followed by mixing and kneading to prepare a dough. Then, boiled, mashed, strained azuki beans in jam (koshi-an in Japanese) is covered with a wrapping prepared from the dough, and the resulting stuffed matter is further rolled with a salted cherry blossom leaf, to prepare a sakura-mochi. The sakura-mochi using as the principal raw material the modified wheat flour can keep softness even after a long time passes and is tasty.

It is needless to say that the preparation of pastas such as spaghetti and macaroni, Japanese noodles such as udon and soba, and Chinese noodles (including the wrappings of gyoza, wang-tang, and the like) may satisfactorily follow routine methods, except that the modified cereal flour of the present invention should be used, in place of conventional cereal flour.

For pasta, for example, water is added to and kneaded with the modified wheat flour and whole egg powder for routine use as a secondary raw material, to prepare pasta dough. The dough is then held at a given temperature (so-called "aging" process), followed by primary rolling, lumping and press rolling, and the resulting dough is finally cut into the

desired width and the desired length, to prepare pasta strips. The pasta strips are boiled in water, together with a small amount of edible salt, and the resulting pasta can attain preferable hardness, excellent crispness and preferable elasticity. This is possibly due to the formation of a network structure in the gluten of wheat owing to the action of transglutaminase.

5 It is needless to say that the modified wheat flour of the present invention is also used for producing noodles other than pasta, such as Japanese noodles including udon and soba, Chinese noodles and the like. The overall production of these noodles can be done according to methods conventionally used, except for the use of the modified cereal flour, instead of conventional cereal flour.

10 **Examples**

The present invention will now be described in detail in the following examples.

Example 1 (Chinese noodle)

15 Attempts were made to prepare a modified wheat flour from a raw material Canadian wheat (Canada Western Red Spring species; 9 kg).

20 Firstly, micro-fine stone, micro-fine iron debris and the like were removed from the raw material wheat (selection process). Then, the thus selected wheat was placed in a tank, to which was added water to the extent that the final water content in wheat was about 14.7 %. Then, the wheat was kept as it was at 25 °C for 24 hours (tempering process). Transglutaminase was previously added to and dissolved in the water to be added, to a final concentration of 5 U per 1 g of the wheat protein.

25 After tempering, the wheat sheath was separated to recover crude grain of the endosperm (disruption process). Then, contaminated sheath debris was removed from the crude grain of the endosperm by means of a combination of sifting and draft selection (purification process). Furthermore, the purified crude grain was ground with a smooth roll and sifted by the size of the grain (pulverization process), to obtain 7 fractions, depending on the size.

Among the 7 fractions thus recovered, wheat flour of the second largest size and wheat flour of the fourth largest size were blended together (blending process), followed by sufficient mixing (finishing process). The modified wheat flour thus obtained was defined as Inventive Product 1.

30 A modified wheat flour obtained by the same milling process except for the addition of 10 U of transglutaminase instead of 5 U thereof per 1 g of the wheat protein at the tempering process was defined as Inventive Product 2. Similarly, a modified wheat flour obtained by a milling process including transglutaminase treatment with addition of 5 U of transglutaminase per 1 g of the wheat protein and addition of 0.1 g of the partial protein hydrolysate of the wheat protein per 1 g of the wheat protein was defined as Inventive Product 3; and a modified wheat flour obtained by a milling process including transglutaminase treatment with addition of 5 U of transglutaminase per 1 g of the wheat protein and addition of 1.0 g of the partial protein hydrolysate of the wheat protein per 1 g of the wheat protein was defined as Inventive Product 4. Herein, as the partial protein hydrolysate, a partially hydrolyzed product of the protein part of wheat was used, with an average molecular weight of about 7,000 (trade name; Glutamine peptide, manufactured by "Canpina Milk Uni-Japan").

40 As a control product, general wheat flour produced without addition of any of transglutaminase and the partial protein hydrolysate, namely general wheat flour produced through a milling process without any such transglutaminase treatment as described above, was used.

See furthermore Table 1 below.

45

Table 1

50

55

Wheat flour	Transglutaminase U/g protein	Glutamine peptide g/g protein
Control Product	0	0
Inventive Product 1	5	0
Inventive Product 2	10	0
Inventive Product 3	5	0.1
Inventive Product 4	5	1.0
g protein; per 1 g of wheat protein		

The four types of modified wheat flour and the control, thus obtained, were individually weighed to obtain samples of each 2000 g, with which were blended edible salt (20 g), Chinese noodle seasoning (kansui; 20 g) and water (800 g), followed by kneading at 500 mmHg by means of a vacuum mixer (vacuum mixer of type TVM 03-0028, manufactured by Tokyo Noodle Machine, Co.) for 10 minutes. Subsequently, the resulting kneaded product was rolled loosely by means of a noodle machine manufactured by Shinagawa Noodle Machine, K.K., followed by lumping twice and press rolling four times and the resulting product was cut, to prepare five types of Chinese noodles.

A sensory evaluation of the Chinese noodle samples by a panel of 10 experts was carried out by a 10-point scoring method, under the provision that the control product be scored as 5 points. The average points are shown in the following Table 2. Additionally, the cutting energy was measured by a cutting test method with a rheometer, and the results are shown in the same table. In the table, furthermore, the representation of individual Chinese noodle types respectively corresponds to the representation of the wheat flours as the raw materials. (For example, the Chinese noodle of the Inventive Product 2 is prepared from the modified cereal flour of the Inventive Product 2.)

Table 2

Chinese noodle	Sensory evaluation			Properties cutting energy (erg/cm ²)
	elasticity	viscosity	limpness	
Control Product	5 points	5 points	5 points	11.2 x 10 ⁴
Inventive Product 1	7.0	7.0	7.1	17.2 x 10 ⁴
Inventive Product 2	6.9	7.6	7.7	17.8 x 10 ⁴
Inventive Product 3	8.5	8.6	8.4	18.6 x 10 ⁴
Inventive Product 4	9.1	9.2	9.0	19.3 x 10 ⁴

Table 2 above indicates that Chinese noodles prepared from any of the modified wheat flours treated by the transglutaminase only as well as treated by a combination of transglutaminase and the partial protein hydrolysate as the raw materials are not only excellent from the sensory respect but also have greater cutting energy from the respect of objective parameter. The data of objective parameter supports that the Chinese noodles obtained by using the modified wheat flours have the most suitable taste as noodles.

Example 2 (A loaf of bread)

Attempts were made to produce four types of modified wheat flours and a control product, in the same manner as in Example 1, except for the use of the partial protein hydrolysate of a wheat protein with an average molecular weight of about 10,000 (trade name; MA-Z, manufactured by Morinaga Milk Industry, Co., Ltd.), instead of the partial protein hydrolysate of a wheat protein with an average molecular weight of about 7,000 (trade name; Glutamine peptide, manufactured by Canpina Milk Uni-Japan).

1400 g each of the four types of the modified wheat flours and the control product, thus obtained, was weighed, followed by addition of yeast (40 g), yeast food (2.5 g) and water (750 g) and subsequent mixing at a low speed for 2 minutes, next a medium speed for 4 minutes and then at a high speed for 1 minute with a Hobart mixer. After that, processed fat (50 g) was added to the resulting dough, followed by another mixing at an intermediate speed for 3 minutes and a high speed for 1 minute. Then, the dough was kept under the condition of 27°C and 75% RH(Relative Humidity) for 4 hours, as first fermentation to obtain intermediate seed dough. The dough on completion of fermentation was at 28 °C and pH 5.3.

To the intermediate seed dough were added other ingredients (40 g of edible salt, 60 g of sugar, 60 g of glucose, 60 g of shortening, 40 g of skimmed dry milk and 440 g of water), followed by kneading with a mixer to prepare bread dough. The bread dough was kept at the above-mentioned temperature for about 10 minutes (second fermentation), which was then divided equally in 6 portions and further kept at 28°C for 10 minutes (third fermentation). Then, the resulting dough was filled in a pan. The dough was kept under conditions of 37°C and 75% RH for 50 minutes for another fermentation (fourth fermentation). Subsequently, the fermented bread dough was placed in an oven, for baking at 220°C for 40 minutes to prepare 5 types of loafs of bread.

According to the following method, the resulting 5 types of loafs of bread were evaluated in an organoleptic manner. More specifically, the loafs of bread 4 days after baking were sliced at a 1.5-cm thickness, and then they were evaluated by a panel of 10 specialists. Additionally, evaluation was carried out by the 10-point method on the basis of the scoring

shown in Table 3 below, wherein individual characteristics of the control product were scored as 5 points. The average points of each result of five evaluation items, i.e. surface color, surface quality, acid expansion, inner color phase and texture, are shown in Table 4 below.

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Table 3

Evaluation standard	10 very strong
	9 fairly strong
	8 strong
	7 relatively strong
	6 slightly strong
	5 normal
	4 slightly weak
	3 relatively weak
	2 weak
	1 fairly weak
	0 very weak

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Table 4

	Surface color	Surface quality	Crumb grain	Inner color phase	Texture
Control Product	5 points	5 points	5 points	5 points	5 points
Inventive Product 1	8.0	7.0	7.5	8.0	7.5
Inventive Product 2	8.5	7.5	7.5	8.5	7.5
Inventive Product 3	8.5	8.5	8.0	8.5	7.5
Inventive Product 4	9.0	9.0	8.5	8.5	7.6

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Table 4 indicates that all the loafs of bread (Inventive Products) prepared by using the modified wheat flours treated by only transglutaminase or a combination of transglutaminase and the partial protein hydrolysate are excellent from the sensory standpoint, compared with the control product.

Example 3 (Sponge cake)

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A modified wheat flour was prepared by using American wheat (Standard White species; 10 kg) as the raw material.

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Firstly, micro-fine stones and the like were removed from the raw material wheat (selection process). Then, the wheat thus selected was placed in a tank, followed by addition of water to the extent that the final water content in the wheat was about 14.3% , and then, the resulting wheat was kept as it was, at 25°C for 36 hours (tempering process). Then, transglutaminase was added to and dissolved in water to be added, at a rate of 1 U per 1 g of the protein of the wheat.

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After tempering, the wheat sheath was separated, to obtain the crude grain of the endosperm (disruption process). Then, contaminated sheath debris was removed from the crude grain of the endosperm by means of a combination of sifting and draft selection (purification process). Furthermore, the purified crude grain was ground with a smooth roll (pulverization process). Subsequently, the resulting flour was sifted, depending on the size of the flour, to obtain 7 fractions, depending on the size.

Among the 7 fractions thus recovered, the wheat flour of the second largest size and the wheat flour of the fourth largest size were blended together (blending process), followed by sufficient mixing (finishing process). The modified wheat flour thus obtained was defined as Inventive Product 1.

By the same milling process, except for the use of 1 U of transglutaminase per 1 g of the wheat protein and 0.1 g of sodium caseinate (manufactured by Nissei Kyoeki, Kabushiki Kaisha) per 1 g of the protein in wheat, Inventive Product 2 was recovered.

Furthermore, see Table 5 below.

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Table 5

	Amount of transglutaminase (U/g protein)	Amount of sodium caseinate(g/g wheat flour)
Control Product	0	0
Inventive Product 1	1	0
Inventive Product 2	1	0.1

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By using the two types of the modified wheat flours and control wheat flour (wheat flour obtained in the same manner except that the transglutaminase treatment was not done), sponge cake of the following composition shown in Table 6 was prepared by routine methods.

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Table 6

Composition of sponge cake	
Raw material	Composition
Modified wheat flour or Control wheat flour	200g
Sugar	200g
Whole egg	180g
Water	85ml

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More specifically, whole egg was placed in a bowl, followed by addition of water (45 ml) and thorough mixing with a Hobart mixer, and to the resulting mixture was added sugar. Then, the mixture was adjusted to 30°C. Subsequently, the mixture was foamed at a high speed for 8 to 10 minutes. After adding the remaining water (40 ml) to the mixture, the mixture was foamed for 2-min at a high speed and subsequently for 1-min at a low speed, thereby cake dough was prepared. The modified wheat flours or the control wheat flour was added to the cake dough, followed by sufficient mixing. The resulting dough was placed in a pan, followed by smoothing the surface by means of a plastic pallet, and the resulting dough was then baked in an oven at 180 °C for 30 minutes. After baking, the product was drawn out from the pan and cooled to room temperature, and then subjected to sensory evaluation by a panel of 10 experts. The evaluation results are shown in Table 7 below. Furthermore, the appearance and inner phase were observed; and the volume of the cake was measured. These results are also shown in the same table. The overall evaluation based on all of these results is also shown in the same table. The symbols X, Δ, ○ and ⊙ (double circle) represent not good, normal, relatively good and very good, respectively.

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Table 7

	Appearance	Cake volume	Inner phase	Texture	Overall
Control Product	recess at center	2400ml	pale yellow	slightly hard	Δ
Inventive Product 1	no recess	2450	white	soft and smooth	⊙
Inventive Product 2	no recess	2480	white	soft and smooth	⊙

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As shown in the above table, sponge cakes (the inventive products) with a larger volume without recess at the center and preferably with soft and smooth texture, were prepared, compared with the control product.

Example 4

By the same procedures as in Example 1, attempts were made to produce 4 types of modified wheat flours and a control product.

The extensograms of the 4 types of the modified wheat flours and the control wheat flour were measured by the following method. Herein, the secondary processing property of wheat flour is measured by such extensogram.

Subsequently, 300 g each of the 4 types of the modified wheat flours and the control wheat flour was placed in a farinogram mixer (trade name: "Farinogram", manufactured by Brabender, Co.), followed by addition of 6 g of edible salt and an appropriate amount of pure water, to prepare dough. More specifically, in the dough preparation the amount of pure water was so adjusted that the peak of the curve of the dough after 1-min kneading, 5-min holding and another 2-min kneading might be 500 B.U.

From the dough thus prepared, 150 g thereof was drawn out and molded in a molding machine, which was then kept in a thermostat at 30 °C for 45 minutes. After that a first measurement of the extensogram was done. Forty-five minutes after another molding (namely 90 minutes after the initiation of 1st. keeping), a second measurement was done. Forty-five minutes after further molding (namely, 135 minutes after the initiation), a third measurement was done. Herein, the measurement was carried out with a machine called "Extensigraph" manufactured by Barbender, Co.

The data 135 minutes later is shown in Table 8, which functions as the optimum indicator of the elasticity of wheat. Herein, R in the table represents tensile resistance, which is the peak height of the curve of an extensogram measured (the unit is B.U.). Additionally, E represents extension, which is the length of the curve of the measured extensogram (the unit is mm). Furthermore, R/E represents shape coefficient, which is a value of tensile resistance divided by extension. A larger R/E represents a higher elasticity of wheat flour.

Table 8

	Control Product	Inventive Product 1	Inventive Product 2	Inventive Product 3	Inventive Product 4
Tensile resistance (R)	278	415	570	525	585
Extension (E)	218	157	180	121	110
Shape coefficient (R/E)	1.28	2.64	3.17	4.34	5.32

As apparent from Table 8, all the Inventive Products were wheat flours having higher elasticity than that of the control product.

Advantages of the Invention

According to the present invention, modified cereal flour with excellent processability can be readily produced, and thus, cereal-flour based processed food of high quality can be readily produced. In accordance with the present invention, furthermore, transglutaminase is added to function during the process of producing wheat flour from wheat as an agricultural product, to readily produce a wheat flour with excellent processability. Therefore, flour industries can readily manufacture and sell such wheat flour of high quality, and thus, the advantages of the present invention for flour industries are enormous.

The modified wheat flour of the present invention has a lower allergen content and is hardly deteriorated under storage for a long term, compared with conventional products. Specifically, transglutaminase treatment in combination with a foreign protein and/or a partial protein hydrolysate causes far less allergy.

Claims

1. A modified cereal flour obtained by effecting a transglutaminase treatment at the following process steps for producing cereal flour from cereal grains as the raw material:
 1. tempering process and/or
 2. grinding process
2. A modified cereal flour according to claim 1, wherein the transglutaminase treatment is carried out in the presence of a foreign protein and/or a partial protein hydrolysate.

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3. A modified cereal flour according to claim 1 or 2, wherein the transglutaminase treatment is carried out by using 0.01 to 100 units of transglutaminase per 1 g of the protein in the cereal grain.
- 5 4. A modified cereal flour according to claim 2 or 3, wherein the foreign protein is wheat protein or milk protein and the partial protein hydrolysate is derived from wheat protein or milk protein.
5. A modified cereal flour according to any one of the claims 1 to 4, wherein the raw material cereal grain is wheat and the modified cereal flour is modified wheat flour.
- 10 6. A cereal-flour based processed food, produced from a modified cereal flour according to any one of the claims 1 to 5 as a raw material.

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